

SKULL SEXUAL DIMORPHISM APPEARS IN TOY RABBITS

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ABSTRACT. The aim of this paper was to use geometric morphometrics to study the skull sexual dimorphism of toy rabbits, which present paedomorphic (babyiness) traits comparing data with those from agriotype (ancestor), *Oryctolagus cuniculus*. For this purpose, we examined 43 post-weaned corpses belonging to wild rabbit (n=22, 7 ♂ and 15 ♀, 1295.6 ± 333.8 kg of body weight and 88.0 ± 1.12 cm of ear length) and toy type (n=21, 4 ♂ and 17 ♀, short and upright ears, 1031.3 ± 644.13 kg of body weight and 6.2 ± 1.08 cm of ear length). Heads were radiographed using a Potro® machine on a latero-lateral projection and 7 landmarks were located on the skull and studied by means of geometric morphometric procedures. Size and shape between genders appeared statistically different only for toys, mainly focused on splanchnocranium (face) for shape. Detected sexual dimorphism could be attributed to selection arising from differential mating success, or sexual selection, due to human management. Moreover, the inconsistency with Rensch's hypothesis – which establishes that males in larger species will tend to be larger relative to females than in smaller species- allows us to suggest that Rensch's hypothesis is not necessarily followed in artificial selection experiencing miniaturization in body shape. It must be outlined the opportunities to tackle paedomorphic questions via geometric morphometrics methods in toy rabbits.

1. INTRODUCTION

Toy rabbits are a type of the wild rabbit (*Oryctolagus cuniculus*) selected by their paedomorphic traits. Toys are typical for *babyiness* traits [1]: relatively big skull, shortened rostrum, and short ears, which render them especially attractive to owners [2]. Under development of a trait relatively to the ontogenetic course of this trait in the ancestor correspond to paedomorphosis [3,4]. Toys underdevelop some traits, giving them a clear infantile aspect. There is some evidence that there are functional constraints represented mainly by miniaturization of size in small toys [2]. Breeders point out that toy females may suffer from complicated parturition. Modern lineages of these types of companion rabbit present a unique opportunity to test hypothesis about paedomorphosis.

The aim of this paper was to use geometric morphometrics to study the skull sexual dimorphism of toy rabbits, comparing data with those from agriotype (ancestor), the wild rabbit, for which, at least from Iberian Peninsula, no dimorphism has been described for

Received by the editors: April 20, 2019; Accepted: December 04, 2019.

Keywords and phrases: *Oryctolagus cuniculus*, neoteny, paedomorphism, progenesis

males and females [5]. Sexual dimorphism for toys and wilds are studied both as size as shape difference.

2. MATERIAL AND METHOD

2.1. Sampling procedure and data collection

The present study examines 43 post-weaned animals belonging to wild rabbit (n=22, 7 ♂ and 15 ♀, 1295.6 ± 333.8 kg of body weight and 88.0 ± 1.12 cm of ear length) and Toy type (n=21, 4 ♂ and 17 ♀, short and upright ears, 1031.3 ± 644.13 kg of body weight and 6.2 ± 1.08 cm of ear length). Fresh corpses of toys were collected from a breeding farm, and wilds were supplied from pest control campaigns. Then, they were beheaded, and ear length was obtained with a calliper. Sampled specimens were sexed whenever possible.

2.2. Data acquisition

Then heads were then radiographed using a Potro® machine on a latero-lateral projection. Exposure values ranged from 40 to 60 kV and 3.2-5 mAs. Pictures (each approximately 1.1 MB) were then saved in jpeg format and transferred to a computer.

2.3. Size and shape analysis

We firstly digitized 7 landmarks (LMs) by TpsDig 2.16 [6] to obtain the x - y coordinates of all points (Figure 1). The landmarks included in this study are chosen to correspond to those commonly used in both traditional [7] and GM. For the same 45 individuals, all images had a double digitalization of all landmarks for assessing the measurement error. First author (PMPC) was responsible of this preliminary study.

To perform the study, LMs were converted to shape coordinates by the generalized least square (GLS) Procrustes superimposition (GPA). GPA preserves all information about shape differences among specimens removing information about location, orientation and rotation from the raw coordinates and standardizes each specimen to unit centroid size (CS, a dimensionless size-measure computed as the square root of the summed squared Euclidean distances from each landmark to the specimen centroid) [8]. The information about the shape variation was extracted from the Procrustes superimposition [9,10]. Then we extracted the covariance matrix, generated by the Procrustes coordinates, and that includes the measures of the association between Procrustes coordinates themselves (that are the X and Y coordinates of each landmark after the Procrustes superimposition) [9,10]. The covariance matrix was used as a base for the subsequent analyses. A Mann-Whitney test was done to analyse CS differences between sexes, while a NPMANOVA using the Euclidean distance was used to study shape differences between types. A *deformation grid* was used to capture

the morphological shape differences and changes. Geometric procedures were performed in MorphoJ version 1.06c [11] and the rest of analysis with PAST version 2.17c softwares [12].

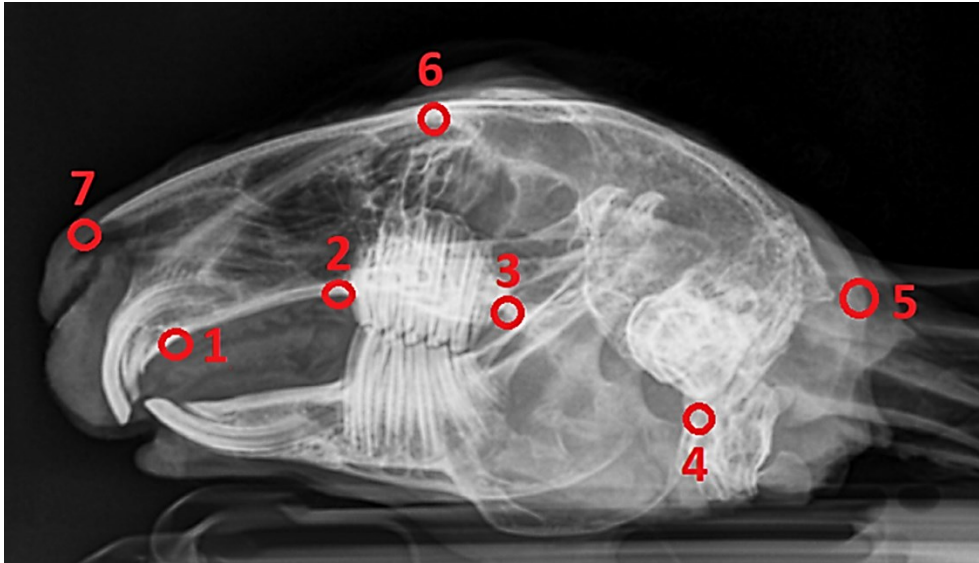


FIGURE 1. Location of the 7 skull landmarks used in the analysis: 1: base of inner upper incisor teeth; 2: most basal oral point of maxillary premolar teeth series (1st pM); 3: most basal caudal point of maxillary molar teeth series (3rd M); 4: ventral point of tympanic bulla; 5: external occipital protuberance; 6: dorsal projection of anterior cranial fossae; 7: most oral point of nasal bone. Latero-lateral projection.

3. RESULTS

Step 1 - Error evaluation

The evaluation of measurement error by the Procrustes analysis of variance (ANOVA) showed that error was negligible ($F=0.04$, $p=1$). Thus, all ulterior analyses went on using the averaged two replicas.

Step 2 - Analysis of size

Mean male and female CS were shown by Mann-Whitney test to be no significantly different for wilds ($U=52$, $p=1.0$, average of 617.84 (s.d. 44.33)), while for toys there appeared

significant differences ($U=10$, $p=0.034$, average of ♂ 476.6 (s.d. 72.99) and ♀ 577.4 (s.d. 80.9)). This indicates skull size sexual dimorphism only in toy rabbits.

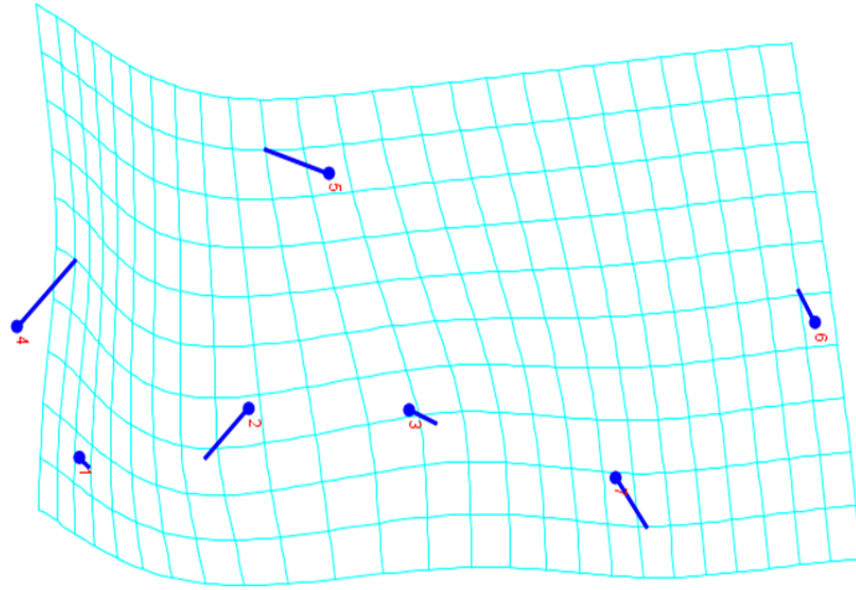


FIGURE 2. The differences were focused on splanchnocranium (face) (7 landmarks; see Figure 1).

Step 3 - Analysis of shape

The NPMANOVA test (Euclidean distances) indicated that the Procrustes distance between sex means was significantly different for skulls of toys ($F=192.3$, $p=0.036$) but not for wilds ($F=0.950$, $p=0.464$). This indicates skull shape sexual dimorphism only in toys skulls. In terms of geometric morphometrics the differences were focused on splanchnocranium (face) (Figure 2).

4. DISCUSSION

Geometric morphometrics provide the opportunity to get new insights in the variety of morphological characteristics and morphs of wild and domestic rabbits. The technique not constrained by focusing on particular shape features a priori, so that it was possible to detect differences in any direction of shape space. Such shape differences among groups can be easily visualized through deformation grids. The positioning of landmarks can be

individually adapted to particular research questions, so that geometric morphometric methods can be broadly applied for a wide variety of morphological questions. Traditional comparative morphological approaches are often based on selected measurements, and results are somewhat restricted to those few variables.

Toy rabbits, exhibiting paedomorphy did not present head sexual dimorphism, while the agriotype (ancestor wild species,) did [5,13]. Probably it reinforces Gould's conviction that fairly simple epigenetic perturbations often underlie complex morphological evolutionary changes [14]. Rensch's hypothesis establishes that males in larger species tend to be larger relative to females than are males in smaller species [15,16], but this was not the case, as toys were smaller than ancestors.

The difference in consistency with Rensch's hypothesis between wild rabbit and toys allows us to suggest that Rensch's hypothesis is not necessarily followed in artificial selection towards a miniaturization. Sexual dimorphism patterns not consistent with Rensch's hypothesis have been demonstrated in domestic chicken breed, too [17], and in fact this is logical if we keep in mind that artificial selection and formation of breeds (or varieties, or lineages) in domesticated animals is a different process involving for instance different genetic changes than speciation [18,19]

The link between developmental processes which suggests that developmental polymorphisms could affect variation in sexual size dimorphism [20] could reinforce this hypothesis. Thus, there are several promising trajectories to address important morphological questions on paedomorphy among domestic mammals, so that there is no doubt that this field will evolve further rapidly.

Acknowledgements. The authors thank CUNIPIC, in Tèrmens (Catalonia) for providing corpses of toy rabbits. Also, we are grateful to anonymous reviewers for their insightful suggestions.

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BMC Evolutionary Biology 9(1): 278.

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